INFLUENCE OF RADIATION STERILIZATION ON RESPIRATION AND OTHER PROPERTIES OF DORMANT WHEAT SEEDS

by

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INTRODUCTION

Moisture content is the major factor responsible for the biological deterioration of wheat in storage at ordinary temperatures since it promotes the growth and metabolic activity of certain common fungi whose spores and mycelia exist within the seed coats of normal grain (Milner, 26). At moisture contents below 14 per cent at ordinary temperatures, fungal activity is virtually absent, but as moisture exceeds this point mold spore germination and mycelial growth begin, accompanied by characteristic respiration increases, as well as heating effects if the grain is in a considerable bulk. Thus with time the respiratory trend of freshly wetted grain is in the form of a microbiological population growth curve. Under the same conditions the respiration of the seed itself is much lower and virtually constant with time (Milner and Geddes, 27).

Traditional means used to control this deterioration is by drying of the grain. In the State of Kansas very few elevators have drying facilities, because normally the wheat crop is harvested in a relatively dry condition. It has been proposed from time to time that alternative procedures to drying might be used to preserve damp grain. Among these would be the use of chemical preservatives to inhibit the microorganisms as well as ionizing radiations to achieve the same purpose.

In recent years, the prospect that waste radioactive fission products will become increasing available has suggested their use for the sterilization treatment of various foods including damp

grains. To evaluate this possibility, measurement of respiration including the determination of carbon dioxide output and respiratory quotient, provides an effective means to indicate fungal and seed respiration, and the value of various preservative treatments for prolonging the storage life of damp grains.

This study deals with the influences of gamma radiation treatment on the respiration and chemical deterioration of wheat stored under conditions comparable to commercial conditions.

HISTORICAL REVIEW

Theory of Radiation Sterilization

The direct and indirect "hit" mechanisms govern the fundamental reactions of radiation chemistry. Weiss (41) explained the direct hit action by the absorption of energy by a biologically active giant molecule AB which was followed by formation of radicals A and B according to the scheme:

$$AB + \text{Radiation} \longrightarrow A + B$$

Either a recombination of the fission products to an original molecule,

$$A + B \longrightarrow AB$$

or a reaction of the fission products with other substances which act as an acceptor say X or Y was assumed to be followed.

$$A + X \longrightarrow AX$$

$$B + Y \longrightarrow BY$$

The above three reactions typically represent the fundamental radiation process occurring in the direct "hit" mechanism.

The indirect "hit" mechanism was postulated to take place in dilute solutions and involves formation of active radiation products of water. Fricke (15) proposed that this mechanism could be interpreted by the indirect action involving "activated water." The further development of this idea that "activated water" was identical with atomic hydrogen and hydroxyl free radicals formed by radiation was due to Weiss (42). These two radicals are known to be very reactive and act as reducing and oxidizing agents as well as to the cleave C-C bonds. Practically all substances present in the water will be attacked by these powerful reagents. In the presence of dissolved oxygen, secondary products of irradiation are usually formed and these are of more importance, for the hydrogen atom can combine with molecular oxygen to form the very reactive O_2H radical,

$$H + O_2 \longrightarrow O_2H$$

followed by the combination of two O_2H free radicals to form hydrogen peroxide and molecular oxygen which enters the process again.

$$O_2H + O_2H \longrightarrow H_2O_2 + O_2$$

The direct and indirect "hit" actions can occur at the same time and in the same system. While there is no doubt both modes of action are valid, the indirect theory offers a wider basis for chemical change.

Types of Radiations

There are at present three basic types of ionizing radiations which have been investigated in food research. These are as

follows:

X-rays. X-rays have relatively great penetration into matter. However, only 3 to 5 per cent of the necessary electron energy goes into the production of x-rays, thus exposure time is relatively long and power cost is high.

Cathode-rays (electron beam). Cathode rays are artificially accelerated electrons or beta particles. Sterilization by electron beam offers the advantage of very high efficiency in that of about 75 per cent of the electron beam energy can be utilized. Moreover, sterilization can be achieved in the range of seconds to minutes. The principal disadvantage of cathode ray is its penetration.

Nuclear Fission Products. The third method is the use of nuclear fission products. Radioactive isotopes have been widely used as gamma-ray emitters in food research. Similar energetic radioactive rays would be emitted by fission products produced in nuclear reactors. This type of radiation is potentially useful because its great penetration ability. Since in the radiation process there occurs only a minute rise in temperature in the sterilized products during the short exposure time (in the order of seconds or minutes), this method of processing is often referred to as "cold sterilization."

Effect of Ionizing Radiation on Insects and Microorganisms

Insects were reported to be relatively sensitive to ionizing radiation. Many investigations have been made on the effects of

radiation on various kinds of insects (Duggar, 13). Bushland and Hopkins (9) investigated the doses which prevent reproduction, or which produce lethal effects. Hassett and Jenkins (19) carried out an experiment using the cobalt 60 isotope as a gamma emitter, on six species of insects troublesome to grain, tobacco, and similar products. They indicate that reproduction is inhibited with 16,000 to 32,000 rep[1] and that 64,400 rep doses are completely lethal. Recently Baker et al. (2) reported that a dose of 250,000 rep kills adults and eggs of the granary weevil immediately after treatment, while 500,000 rep had the same effect on the flour beetle, Tribolium. Nickerson et al. (33) have pointed out that irradiation with cathode rays may soon become an economical method for destroying insects in grain products and that this irradiation may be applied to insects in all stages of development. Nickerson et al. (33) further suggested that the destruction of microorganisms by cathode rays follows a first order reaction, and indicated, that in general, the lower the order in the plant and in animal kingdoms, the greater the resistance of the microorganism to cathode rays. This is illustrated in Fig. 1.

Effect of Ionizing Radiations on Various Foods

Numerous investigators have applied ionizing radiations to fruits, vegetables, meats, and other foods. Sterilization involves introduction of sufficient radiant energy into foods to

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[1] Rentgen equivalent physical: one rep is that amount of radiation resulting in the dissipation of 83 ergs of energy per gram of tissue exposed.

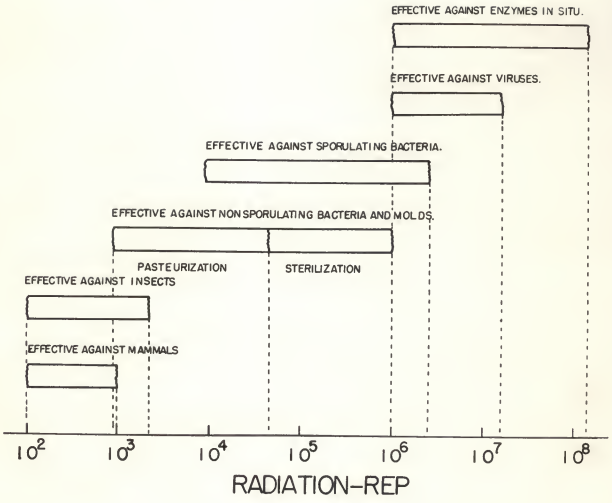


Fig. 1. Dose levels of ionizing radiations required for various biological effects.

destroy all microorganisms. Killing of these microorganisms actually follows from an induced change in their chemical structure, and since radiation cannot be controlled to strike only the microorganisms, some food constituents will be affected. The change in the food chemical components due to radiation are reported to be in the order of only 0.003 per cent of all compounds present, but we must recognize the fact that even at this low concentration some foods appear to be satisfactory while others change in acceptability.

A report (Morgan, 31) on work by Brownell indicated that apple juice after treatment with 500,000 rep showed no loss in color or odor. Irradiation of bananas in general reduces the rate of initial softening but also accelerates skin-darkening. Grapefruit was reported to suffer no loss of acceptability at 500,000 rep. On the other hand lemons lost acceptability below 500,000 rep. Bellamy and Lawton (3) found that potatoes with a 25,000 dose (range of sprout inhibitation) showed inactivation of the enzyme catalase but when mashed potatoes were analyzed for catalase, a dose of 5,000,000 rep was needed for inactivation. Sparrow et al. (40) pointed out that gamma radiation particularly from the range of 5,000 to 20,000 rep can greatly prolong the storage life of potatoes. Effect on taste, sprouting, shrinkage, internal spotting and texture were observed for 18 months. There was no undesirable taste in any irradiated samples after 18 months storage. At the same time, the weight loss had reached 55 per cent in the control (non-irradiation) but considerably less in all irradiated samples with only 20 per cent in the 20,000 rep sample.

Interest in irradiation of meat products has not only been intense but extensively reported (Morgan, 32). Huber et al. (20) pointed out that beef appears to be more stable than veal, and pork the least. Morgan (31) reported Brownell et al's. work that raw ground beef was acceptable after 10 days of storage at ordinary temperatures when exposed to between 50,000 and 100,000 rep.

Proctor et al. (37) found that haddock fillets kept 15 days after a 1,500,000 rep dose showed insignificant increases in trimethylamine and volatile acids and changes in acceptability. In a later study Nickerson et al. (34) reported that haddock fillets irradiated at 700,000 rep and held six weeks at 38° F. were acceptable.

Many investigators in this field have shown that milk can be pasteurized by comparatively low doses of high voltage cathode rays. A report (Morgan, 31) on work by O'meara indicated that even at 100,000 rep given at 50 to 60° F., an undesirable flavor was noticed. This adverse flavor decreased slightly on storage, and was minimized by irradiation in the frozen state. Little success was achieved with concentrated milk and dried milk.

Effect of Ionizing Radiations on Proteins

Proteins are large molecular aggregates of amino acids joined through peptide bonds, with molecular weights ranging between 34,000 and 200,000. The amino acids residues are believed to be oriented in definite patterns within the structure of the

molecules and to be held in these patterns by hydrogen bonds and secondary valence forces between numerous polar functional groups. The most obvious effect of ionizing radiations upon protein is the denaturation of the protein molecule. Fricke (16) pointed out that the stability of protein with respect to the denaturation is lowered upon treating with ionizing radiation as evidenced by the fact that the stability of egg albumin towards denaturation was lowered by X-irradiation prior to heating. In a later more theoretical study Fricke (17) using both irradiated and non-irradiated egg albumin solutions, he calculated the energy of activation "E" from the measurement of the first order velocity constant of the heat denaturation reaction. He found that "E" was much lower in the case of irradiated solution and concluded that "the irradiation protein is initially undenaturated but contains relatively large quantities of molecular species of lower thermal stability characterized by much lower values of energy of 'activation.'" He explained the lowering of "E" value by suggesting the breaking of side chain bonds or in extreme cases where very strong dosages of irradiations are used, the breaking of primary bonds in the polypeptide chain.

Clark (11) suggested that the denaturation occurred in three steps: 1. An alteration of the protein molecule by the radiation, 2. A reaction between the radiation-altered molecule and water, and 3. The formation of a visible coagulum.

Pauling et al. (35) reported that the energy of formation of the hydrogen bonds is of the order of 8 Kcal. per mole indicating a relatively unstable bond when contrasted to the energy of

formation of covalent bonds. Thus it would be expected that the internal structures of labile molecules or protein would be disrupted by high frequency radiations which supply energies in the order of 1,000,000 Kcal. per mole. In a recent study of effects of x-rays on wheat gluten, Lloyd (22), using viscosity as the principal criterion of effects, indicated that the relation between dosage and the loss of viscosity of sols prepared from irradiated dry gluten was exponential, indicating that separate mechanisms were responsible in each case. Direct irradiation was more efficient in decomposing the gluten in solution, indicating that part of the energy absorbed by the water was transferred to the solute. He further reported that the gluten decomposition was increased as the solute concentration was lowered. This phenomenon supports the proposed activated solvent theory of the action of ionizing radiation of solution which was postulated to be more efficient for dilute solutions. He reported that the proteolytic enzyme associated with gluten in its preparation was not observably inhibited by x-radiation (200,000 rep) of the lyophilized or dissolved preparations. A solution of gluten purified by ether-extraction and dialysis was reported by him to be less sensitive to x-ray reduction of viscosity than an unpurified sol when given doses of 200,000 and 400,000 rep.

Effect of Ionizing Radiations on Wheat and Other Seeds

The effects of ionizing radiations have been recorded for many species of seeds. When applied to wheat (Bless, 6), tomatoes (MacArthur, 23), and many beans (Quastler and Baer, 38) excessive

irradiation resulted in injury manifested as reduced or delayed germination, inhibition of growth, and in some instances, complete loss of viability. Similar unfavorable reactions were observed when cottonseed was irradiated with X-rays (Moore and Haskins, 30). Benedict and Kersten (4) determined the values of wheat sprouted between blotters and the diastase activity, the reducing sugar content, the respiratory rate, and the percentage of water in the seedling after wheat seed was irradiated with the characteristic K lines of copper for various lengths of times. The authors found that wheat irradiated for five seconds showed an increase in both diastase activity and sugar content. But those irradiated for a longer time showed a decided and progressive decrease in these two substances as well as a decrease in the other quantities determined. Afanaseva (1) reported that the action of strong doses (8,000 to 10,000 rep.) of X-rays on the wheat grain brings about through cellular change, a strong stimulating effect which depends not only on the doses of X-rays but also on the condition of the seed. The air-dried seed is least, the moist grain more strongly, and the germinated grain most strongly stimulated by the radiation. He further indicated that moistening immediately before radiation increases the sensitivity only toward the strong doses of X-rays. For weaker doses, a six hour soaking causes no increase in sensitivity, but the germinated grain reacts very strongly even to weak doses (1,000 and 2,000 rep) which produce practically no action on the moist or dry grain. Maes and Bauwen (24) studied the effect of infrared radiation on the keeping qualities of wheat germ. Acid determination was followed during the

storage after the samples were given radiation. The authors found that the amount of acid increases during storage and the increase is less after longer irradiation and if the exposure layer is thinner, but the effect is less pronounced if the germs are milled. The authors also found the same effect with palm kernels, and concluded that infrared irradiation might retard the lipolytic activity.

Lambou et al. (21) studied the adverse effect of high-voltage cathode rays on germination and growth of cottonseed when these rays are applied in dosages of sufficient strength to destroy associated microorganisms. They found that a dose of 1,500,000 rep inactivated the microflora in the seeds. The viability decreased regularly as the dosage of ionizing radiation increased from 500,000 to 1,500,000 rep. Irradiation with 2,000,000 rep destroyed viability in all the seeds. The authors also indicated that these treatments caused no changes in the moisture and fatty acids content of the seed.

Studies on Flour and Bread

Brownell et al. (8) reported that cake flour (Swansdown), all purpose flour (General Mills), and bread flour were not changed when given a dose of 20,000 rep gamma radiation but that flour insects and weevils would be destroyed and insect eggs made sterile. Cake made with these different flours given this radiation dosage were similar to the control in all respects. Cakes made with irradiated bread flour were progressively of lower quality with increasing doses of gamma radiation, and the cakes had low total

volume, were heavier, more compact, gummier, and had a darker yellow crumb color. The authors also pointed out that the gluten appears to lose its binding power and the flour has a drier or more starch-like texture in mixing. Flavor damage was noticed in cakes made with bread flour given a dose of 100,000 rep. Off-flavors were noticed with higher doses of irradiation.

Biscuits made with irradiated flour had a satisfactory appearance but the eating qualities and flavor were not equal to those of the controls. Biscuits were gummier and had off-flavors when the flour was given a dose of more than 200,000 rep.

The authors also observed that in bread produced from flour treated with 200,000 rep the total volume was slightly smaller but that the loaves were equal in all other respects to those made with the non-irradiated flour. The loaves reported to have smaller volume and crumb color were comparable to honey bread when the flour was given doses of 5×10^5 and 10^6 rep.

Summary of the Literature

The literature dealing with irradiation of food is very extensive and is concerned primarily with fresh foods with high moisture content such as fruits, vegetables, and meats. Little pertinent information exists on the effect of irradiation upon dormant seeds such as ordinary wheat in storage at dosages beyond those required to destroy insects (approximately 100,000 rep). Information is needed on the effect of radiation treatment on respiration and storage changes due to fungi and other microorganisms. The effect of such dosages on the technological

properties (milling and baking characteristics) is also not known.

STATEMENT OF THE PROBLEM

In the present study, sound wheat of high quality was irradiated to various dosage levels in both dry and damp conditions. Prevention of deterioration of damp grain prior to irradiation was accomplished by shipment to the radiation facility at Brookhaven National Laboratory after freezing and maintaining the damp grain in dry ice. Studies of respiratory characteristics of treated and untreated grain including carbon dioxide evolution, oxygen uptake, and respiratory quotients were carried out. Seed viability, fatty acid production, development of fluorescence, and alteration of protein solubility were also determined. The investigation was completed with a study of the effect of various dosages of ionizing radiation on the milling and baking properties of the wheat.

MATERIALS AND METHODS

Wheat

A sound sample of hard red winter wheat, grown at the Agronomy farm at Kansas State College in 1954 was used in this investigation. This sample was characterized as follows:

Variety	Concho
Test weight (lb. per bu.)	62.6
Moisture content (%)	8.3
Germination (%)	96.0
Protein content (%)	14.0

This wheat was stored in a cold room at a temperature of 50° F. during the period of this study.

Sample Containers Used for Radiation and Respiration Study

The dimensional restrictions of the radiation facility at Brookhaven National Laboratory were such that the maximum size of the sample container was limited to 1 3/4 inches maximum outside diameter. For this reason, six containers were fabricated from soft 18 gauge aluminum tube with an internal length of 12 1/2 inches and an outside diameter of 1 3/4 inches. In order that wheat samples which had been sterilized by radiation should not become recontaminated with microorganisms, sterile techniques were used. Both ends of each tube through which the air passed were plugged with brass nipples packed with glass usol. The containers were made air-tight by applying non-hardening gasket and joint sealing compound in the threads of the fittings at both ends. After receipt of the irradiated samples, the two small steel plugs on the ends of each container were replaced with two small brass fittings, and to each of these, a piece of copper tube was attached.

A diagrammatic sketch of the sample container used in this

study is shown in Fig. 2.

Shipment of Samples in Dry Ice

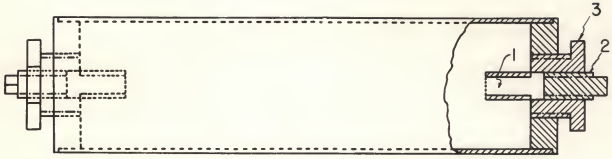
In order to prevent the deterioration and molding of damp grain of 20 per cent moisture content during shipment to Brookhaven National Laboratory and back to Manhattan over a period of eight days, it was necessary to ship the damp grain packed in dry ice. Usually the shipment was made by air express. On receipt of the treated samples in Manhattan, they were unpacked and placed in the constant temperature respiration apparatus, and aeration started immediately.

Irradiation Treatment of Dry Samples

To determine the effectiveness of the sterilization of dry wheat by ionizing radiation, upon return from Brookhaven, sterile water was added to each sample to bring the grain to approximately 20 per cent moisture content, on the basis of the amount of dry wheat in the sample container. The use of sterile technique and the presence of tightly-packed glass wool air filters in the respiration tubes, resulted in completely successful wetting of the irradiated grain without introduction of microorganisms. The grain wetted in this manner was well mixed and stored overnight in the cold room before placing in the respiration apparatus.

Measurement of Respiration

A number of methods have been used to study the respiration



1. Brass nipple packed with glass wool
2. Steel plug
3. Copper bushing.

Fig. 2. Sample container for radiation and respiration study.

of seeds on a laboratory scale. The various types of apparatus employed generally may be divided into two categories, i. e., closed and aerated system. In this study, respiratory rates and respiratory quotients exhibited by this wheat under various conditions were determined using the continuous aeration technique described by Milner and Geddes (29). This apparatus can accommodate six samples simultaneously. The seed samples, maintained in the respirometer tubes in an air thermostat at 30-31° C. were aspired continuously with incoming carbon dioxide-free air (two liters per day) at a relative humidity which was in hygroscopic equilibrium with the initial moisture content of the samples. The air humidification was accomplished by proper concentrations of sulfuric acid. The effluent air accumulated in the spirometers was measured and sampled at 24-hour intervals, and was analyzed for carbon dioxide and oxygen content with a Haldane-Henderson gas analyzer by the technic outlined by Peters and Van Slyke (36). Data were then obtained for the oxygen and carbon dioxide content of effluent air, the respiratory quotient (after applying a nitrogen correction as outlined by Best and Taylor [5], and the total carbon dioxide production.

Measurement of Fluorescence

Cole and Milner (12) indicated that a fluorescence test may be useful for evaluating quantitatively the sick or germ-damaged content of commercial wheat samples, and the test also appeared to be useful as an indication of the viability of such grain. They confirmed the suggestion by Milner et al. (28) that the

discoloration of the germ is due to a reaction of the Maillard type involving the condensation of reducing sugars and proteins which may be a fundamental cause of germ damage in wheat. For the fluorescence measurement, the procedure originated by Cole and Milner was followed. The wheat sample was ground to pass through the No. 30 screen of the intermediate Wilely Mill. A sample size of 2.0 g was used throughout this study. The samples were extracted for 45 minutes in non-stoppered 100 ml Erlenmeyer flasks with 15 ml of 0.186 N. hydrochloric acid, with swirling every 15 minutes. At the end of the extraction, the mixture was transferred to a 15 ml centrifuge tube, and a separation was secured by centrifuging at a speed of 1500 r.p.m. for five minutes. The clear extract was filtered through a No. 4 Whatman filter paper. Clarification of extract was accomplished by adding 5 ml of chloroform and shaking vigorously for one minute. The resulting mixture was centrifuged again for 15 minutes or longer if necessary. In this study, 2 ml of the resulting clear supernatant was transferred to a 50 ml volumetric flask and diluted to volume with hydrochloric acid as used for extraction. Fluorescence measurements were made with Coleman Electron Photofluorimeter with vitamin B_1-S and PC-1 filters transmitting at 345 mμ. Sodium fluorscein (0.075 P.P.M.) was used to standardize this instrument, the dial being set at 60 with this solution.

Measurement of Protein Solubility

The method used for protein solubility measurement, based on per cent transmittancy of potassium sulfate extracts of ground

whole wheat, follows the original method of Zeleny (43) which has been modified by Borensztayn (7). The detailed procedure was as follows: As in the fluorescence measurement, a 2.0 g sample of wheat which had been ground to pass through a No. 30 screen was extracted immediately in a 250 ml glass-stoppered Erlenmeyer flask with 50 ml of 5 per cent potassium sulfate solution for 15 minutes with swirling every five minutes. After filtering the mixture through a No. 4 Whatman filter paper, 10 ml of the filtrate were pipetted into a Coleman Spectrophotometer tube containing 1.2 ml of hydrochloric acid--sodium citrate buffer solution of PH 1.7. The resulting turbid suspension was allowed to stand for 20 minutes. The per cent transmittancy was then read on a Coleman Spectrophotometer at a wave length of 550 mμ.

Sedimentation Test

The sedimentation test is used as a preliminary evaluation of physical qualities of wheat gluten protein. Flour samples used in this test were prepared by a milling micromethod (Finney and Yamazahi, 14). All the wheat samples were conditioned to approximately 14 per cent moisture content and after thorough mixing, were stored in the cold room overnight before milling. The sedimentation test procedure outlined by Grain Division, Agricultural Marketing Services of United States Department of Agriculture, was followed exactly.

Formula and Method Used in Making Bread with Flours Prepared from Non-irradiated and Irradiated Wheat

The following formula and method were used in this bread

baking test.

Formula:

Ingredient	Weight (g)	Weight (%)
Flour	700	100
Sugar	42	6
Salt	14	2
Dry skim milk	21	3
Malted wheat flour	3.5	0.5
Shortening	21	3
Yeast	14	2
$NH_4H_2PO_4$	0.7	0.1
$KBrO_3$	0.000, 0.007, 0.021	0.000, 0.001, 0.003
Water to required absorption		

Straight Dough Method. All ingredients except yeast and water are placed in the mixing bowl. A solution of 200 ml water and the yeast is added to the dry ingredients together with the balance of the required water. The entire mass is then mixed until the dough is developed. After mixing, the dough is placed in fermentation bowls in a cabinet under controlled temperature and humidity (86° F. and 86% relative humidity). After one hour and 50 minutes the gas is punched out of the dough and the dough is allowed to ferment another 50 minutes. Then the dough is scaled to 18-ounce pieces and allowed to rest for 15 minutes at room temperature and humidity. The dough is then moulded by machine and placed in pans which are then placed in a proofing cabinet maintained at a temperature of 95° F. and 95 per cent relative humidity for a period of approximately 50 minutes. Then the pans are transferred to an oven for baking at a temperature of 425° F. for 29 minutes. The volume of duplicate loaves was determined by a standard rape seed volumetric device immediately after the bread was taken from the oven. Loaf properties were evaluated and

overall loaf score was assigned as follows:

Loaf characteristic	Maximum score
Volume	20
Break and shred	5
Crumb color	5
Grain and texture	35
Absorption	10
Dough handling properties	15
Mixing tolerance	10
Total score	100

Miscellaneous Analytical Methods

Moisture. For samples which had been conditioned to moisture content in excess of 14 per cent, the moisture determination was carried out by the two stage air-oven procedure described in detail in Cereal Laboratory Methods (10). For samples of moisture content less than 14 per cent, only the second oven-drying stage of the above method was used. Usually sample weights of from 30 to 40 grams were used for the first stage of air-drying, and the subsequent air-oven stage as well as the various chemical analyses were carried on the air-dried samples.

Fat Acidity. Five grams of wheat which had been ground to pass a No. 30 screen were immediately extracted with about 45 ml of petroleum ether (Skellysolve "B") for six hours in the Goldfisch extraction apparatus. At the end of extraction, the petroleum ether was driven off, and the fat residue was then dissolved by adding 50 ml of 1:1 mixture of 95 per cent ethyl alcohol and benzene containing 0.02 per cent phenolphthalen, and titrated with standard KOH of 0.0178 N. Fat acidity is expressed in terms

of milligrams of KOH required to neutralize the fat from 100 grams of ground whole wheat (in the case of KOH concentration used, 1 ml of KOH was equivalent to 1 mg of KOH).

Germination. All germination tests were carried out by Kansas State Seed Laboratory, at Topeka, Kansas, by the routine methods.

Conditioning Wheat to Various Moisture Levels. Direct addition of water was used to bring all the samples to various desired moisture levels, and the results showed that the distribution of water was very uniform, usually with less than 0.2 per cent variation.

EXPERIMENTAL AND RESULTS

Effect of Gamma Radiation on Respiration, Germination, and Chemical Changes in Wheat Irradiated at 19.8 Per Cent Moisture Content

A lot of wheat was conditioned to 19.8 per cent moisture content overnight, and divided into 11 samples of 250 grams each. These samples were placed in sealed cylindrical aluminum containers and then in frozen storage for one day, followed by packing in dry ice in an insulated container, and shipping by railway express to the Brookhaven National Laboratory at Upton, Long Island. One untreated sample was used as a control and the five remaining pairs were irradiated at increasing dosages of gamma rays as follows:

Sample container	Radiation dosage rep	Radiation time		
		Hours	Minutes	Seconds
A-1	Control (0)	--	--	--
A-2	25,000	--	4	8
A-3	125,000	--	20	40
A-4	625,000	1	43	20
A-5	1,875,000	5	53	0
A-6	3,750,000	11	46	0

The control sample was shipped together with the others in order that it be exposed to the same conditions except for irradiation. Following this treatment these samples were repacked in dry ice and returned by air express. On arrival in Manhattan, one set of samples (A-1 to A-6) was placed in the respiration apparatus. (The remaining set was opened and immediately dried with a fan at room temperature. Chemical analysis was performed on these air-dried samples.) The aeration was started at a rate of 2 liters per day for each sample. Until the samples approached the temperature of the respiration apparatus (30-31° C.), and in order to remove possible accumulation of carbon dioxide produced during the shipment and irradiation interval, 2 liters of air were passed through the sample container. At the termination of the 17-day respiration trial, all containers were opened and the grain immediately air-dried with a fan at room temperature.

The respiratory data obtained with these samples over a 17-day interval at 30-31° C. are given in Table 1, and graphical presentation of the data showing the influence of dosage of gamma

Table 1. Influence of gamma radiation and time on respiratory rate[1] and respiratory quotient[2] of wheat irradiated with moisture content of 19.8 per cent.

	Sample No.											
	A-1		A-2		A-3		A-4		A-5		A-6	
	Irradiation dosages rep											
	Control (0)		25,000		125,000		625,000		1,875,000		3,750,000	
Days	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.
1	48.7	--	50.7	--	--	--	58.9	--	55.8	--	74.9	--
2	31.7	0.92	36.2	0.96	--	--	41.7	0.98	51.2	1.00	49.5	1.36
3	27.5	0.87	33.0	0.97	--	--	35.8	0.90	43.9	1.15	44.5	1.33
4	27.2	0.87	32.6	0.95	--	--	34.0	0.93	41.9	1.57	41.5	1.63
5	28.4	0.95	32.2	0.94	--	--	31.5	1.00	37.0	1.16	36.0	1.66
6	34.7	0.88	37.1	0.98	36.8	1.08	30.3	0.99	33.8	1.24	32.7	1.70
7	52.4	0.93	54.8	0.97	43.1	1.00	30.6	0.97	32.0	1.50	31.3	2.31
8	70.3	0.92	77.2	0.97	59.0	1.00	30.2	1.00	30.0	1.18	28.4	2.04
9	80.8	0.93	92.4	0.95	75.5	1.02	30.0	1.00	29.0	1.30	25.6	2.47
10	88.7	0.86	98.0	0.93	87.6	0.99	29.2	0.97	27.2	1.73	22.0	2.32
11	93.0	0.85	98.7	0.88	91.0	0.96	28.0	--	25.3	1.77	20.0	--
12	96.7	0.81	98.0	0.85	93.0	0.93	27.3	1.02	24.8	1.98	18.0	2.40
13	99.6	0.80	94.2	0.83	92.3	0.87	27.5	1.01	23.6	2.20	14.2	1.74
14	96.0	0.80	91.0	0.83	89.5	0.87	26.8	1.12	22.7	2.40	13.0	2.03
15	91.0	0.80	88.0	0.83	86.0	0.86	25.4	1.04	20.5	2.24	10.3	--
16	88.6	0.81	85.5	0.84	83.0	0.87	25.0	1.15	19.0	2.28	9.1	2.36
17	86.0	0.81	83.7	0.81	80.0	0.87	24.6	1.16	16.9	2.07	8.1	1.79

[1] Respiratory rate in mg CO_2 per 100 g dry matter per day.

[2] Respiratory quotient: Ratio of carbon dioxide output to oxygen consumed.

radiation on daily respiratory rate and respiratory quotient appear in Figs. 3 and 4.

The data in Fig. 3 indicate that a clean-cut elimination of fungal activity was achieved in samples A-4, A-5, and A-6 corresponding to gamma ray dosage of 625,000 rep and higher. The respiration of samples A-1 (control), A-2, and A-3 showed an initial decrease for several days followed by a reversal in this trend. The secondary increase in respiratory activity noted is typical for fungal growth. All of these three samples were obviously heavily molded at the end of the trial. The major species of fungi prevailing on these samples were probably Aspergillus flavus and Aspergillus glaucus (39). Samples A-4, A-5, and A-6 showed a similar decrease in carbon dioxide production but from initially higher levels, related roughly to the degree of radiation dosage. In these samples, however, the decreasing rate of respiration continued and the gas exchange approached zero carbon dioxide production in an asymptotic manner.

Figure 4 shows that the respiratory quotients of samples A-1, A-2, and A-3 were in the range of 0.8 to 1.0 which previous studies have shown to be characteristic of fungal activity. On the other hand the respiratory activity of samples A-4, A-5, and A-6 was accompanied by markedly elevated respiratory quotient values between 0.9 to 2.5. Fungal activity was obviously absent in these three samples.

Table 2 presents analytical data for this series of samples. Gamma irradiation alone has no significant effect on fat acidity.

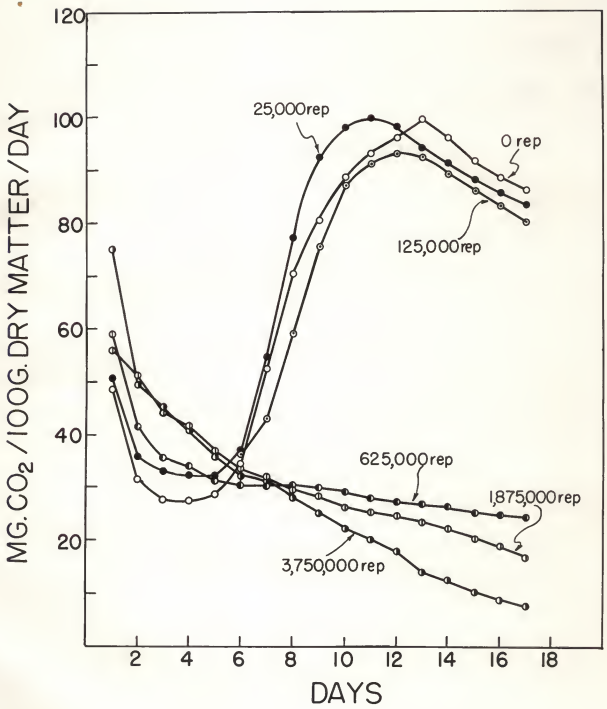


Fig. 3. Influence of gamma radiation and time on the respiration of wheat irradiated at 19.8 per cent moisture.

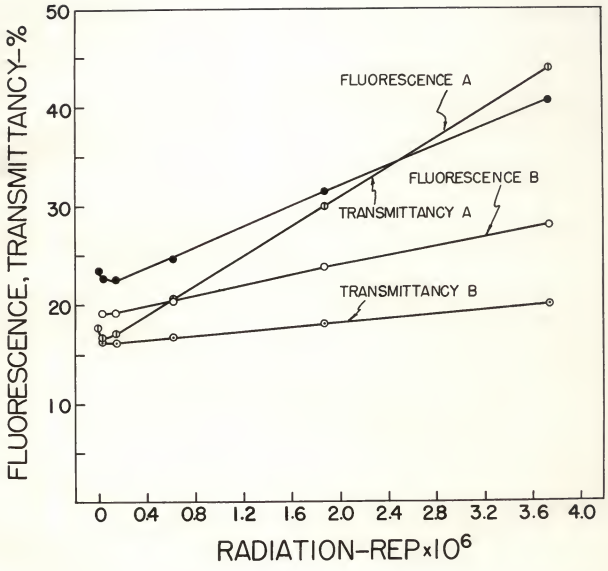


Fig. 4. Influence of gamma radiation on fluorescence and per cent transmittancy (protein solubility of extracts of wheat before (B) and after respiration trial (A) of 17 days at 30-31° C.

Table 2. Influence of gamma radiation on germination, fat acidity, fluorescence, transmittancy (protein solubility), sedimentation value, and moisture content of A series before and after the respiratory trial for 17 days at 30-31° C.

Sample No.	Radiation dosage rep	Moisture content %		Germination %		Fat acidity MgKOH/100g		Fluorescence: scale units		Transmittancy %		Sedimentation value[1]	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
A-1	Control (0)	19.8	20.0	--	7.0	--	53.0	--	23.6	--	17.9	--	45.9
A-2	25,000	19.8	19.9	88.0	2.0	16.4	57.0	19.3	22.8	16.9	16.9	41.6	44.0
A-3	125,000	19.8	19.9	10.0	0.0	16.4	54.0	19.1	22.8	16.3	17.2	42.0	45.9
A-4	625,000	19.8	19.8	0.0	0.0	16.4	17.0	21.0	24.8	16.9	21.1	41.2	45.9
A-5	1,875,000	19.8	19.8	0.0	0.0	15.6	16.0	24.0	31.4	18.1	30.0	41.2	45.9
A-6	3,750,000	19.8	19.5	0.0	0.0	15.6	16.0	28.0	40.4	19.6	44.3	46.8	*
0[2]		8.3	--	96.0		14.0		22.6		14.3		53.7	

[1] 14 per cent moisture basis.

[2] Original sample moisture content 8.3 per cent wet basis.

* No boundary visible.

At the end of respiration trial however, the fat acidities of samples A-1, A-2, and A-3 were considerably higher than those of A-4, A-5, and A-6. Fat acidity has been regarded a sensitive index grain deterioration (Zeleny, 44). The germinability of wheat seeds irradiated at 19.8 per cent moisture content and subjected to eight days of dry ice treatment, decreased very markedly as the dosage of ionizing radiation increased from 25,000 to 125,000 rep. Irradiation with 625,000 rep destroyed viability in all the seeds. The germinability of the samples after respiratory trial was much lowered; taking A-2 for example, it had 88 per cent germination before the respiration trial but it was reduced to 2 per cent afterwards. The combined effect of dry ice treatment, storage in respirometer tube (involving growth of mold), and a dose of 125,000 rep completely destroyed the seed viability.

It can be seen from Fig. 5 that no significant effect was observed on the development of fluorescence of samples A-2, and A-3 following the irradiation. However, it increased regularly as dosage increased from 625,000 to 3,750,000 rep after a 17-day storage interval in respirometers. Fluorescence showed a little increase in samples A-1, A-2, and A-3 in which radiation dosage was insufficient to eliminate the fungi, whereas in samples A-4, A-5, and A-6 in which no fungal respiration was evident, considerable change as manifested by fluorescence apparently occurred. Protein solubility as indicated by the transmittancy values, however, increased slightly with radiation dosage alone up to 625,000 rep and decreased at higher dosage values. Storage in respirometer tube for 17 days after irradiation resulted in little

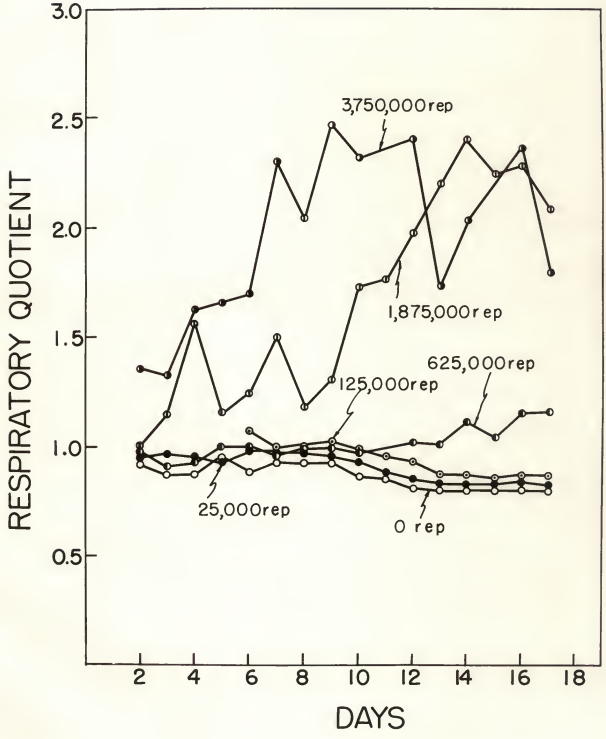


Fig. 5. Influence of gamma radiation and time on the respiratory quotient of wheat irradiated at 19.8 per cent moisture.

change in protein solubility in samples A-1, A-2, and A-3 (in which mold growth occurred) whereas a remarkable decrease in protein solubility was noted in samples A-4, A-5, and A-6 where no mold growth occurred. Sedimentation value, which is an indication of the hydration potential of the wheat gluten, showed little variation in samples A-2, A-3, A-4, and A-5, but increased in A-6 which received the highest dosage. Irradiation alone obviously lowered the sedimentation value below that of the dry untreated wheat. However, it was noted that the sedimentation value of each treated sample after respiration trial is higher than that before respiration trial except for sample A-6 in which no distinct boundary could be observed. No significant variation of moisture content of each sample before and after respiration trial was observed and thus it seems that the relative humidity of the humidifying solution was in equilibrium with the moisture of the grain itself in the respirometer tube.

Effect of Dry Ice Treatment on Respiration, Germination, and Chemical Properties of Wheat

In the experiment with A series, the wheat samples were shipped to the Brookhaven National Laboratory with a moisture content of 19.8 per cent, packed in dry ice. These samples remained frozen for a period of eight days during handling and shipment prior to the initiation of respiration measurement. In order to evaluate the effect of gamma irradiation alone on wheat respiration, it appeared necessary to determine the effect of the preliminary dry ice treatment on the respiratory characteristics of

wheat at 19.8 per cent moisture content. Two sets of six samples (sample weight 250 grams) were prepared. This damp grain (moisture 20.1 per cent) was stored in sealed containers in dry ice for various periods as follows. The grain temperature was recorded as -65° C. during the treatment.

Sample	Time in dry ice at -65° C. (hrs.)
B-1	0
B-2	89
B-3	113
B-4	137
B-5	161
B-6	185

A summary of the data for the daily carbon dioxide production and corresponding respiratory quotient over a period of 21 days at 30-31° C. is given in Table 3. Figure 6 shows the influence of dry ice treatment on the respiration rate. It is evident that in general, the longer the period of dry ice treatment, the lower the carbon dioxide production. There was no initial decrease in carbon dioxide production in samples B-1, B-2, and B-3 but there was some initial decrease in B-4 and B-6. The initial level of carbon dioxide production was highest in B-6 which received the longest period of dry ice treatment, and B-4 was second. The data on B-5 were not completed because of an accident. The increase in respiratory activity in all samples is typical of fungal growth. All of these samples at the end of the respiration trial were heavily molded. The respiratory quotients for each sample were characteristic of fungal activity.

The analytical results obtained with these samples appear in

Table 3. Influence of dry ice treatment and time on the respiratory rate[1] and respiratory quotient[2] at 30-31° C. of wheat with initial moisture content of 20.1 per cent.

Days	Sample No. B-1 (0 hrs.) CO_2	B-1 R.Q.	B-2 (89 hrs.) CO_2	B-2 R.Q.	B-3 (113 hrs.) CO_2	B-3 R.Q.	B-4 (137 hrs.) CO_2	B-4 R.Q.	B-6 (185 hrs.) CO_2	B-6 R.Q.
1	26.6	0.82	48.7	1.34	46.1	1.23	59.1	1.58	103.3	2.60
2	37.6	0.94	57.1	0.97	83.3	0.93	34.2	0.98	37.3	1.03
3	63.7	0.92	83.4	0.93	87.2	0.97	37.5	0.94	39.2	1.07
4	74.0	0.96	106.5	0.96	105.2	1.00	74.6	0.96	55.8	0.96
5	93.0	0.94	112.0	1.00	124.0	0.97	96.0	1.00	88.0	0.97
6	122.7	0.91	120.8	0.93	141.6	0.93	107.7	0.96	96.2	1.01
7	140.0	0.85	--	0.88	152.8	0.83	120.0	0.90	102.0	0.95
8	158.5	0.79	136.0	0.84	163.0	0.80	128.4	0.85	106.8	1.10
9	171.0	0.79	141.7	0.83	165.1	0.81	134.0	0.84	113.0	0.87
10	179.5	0.78	148.0	0.82	165.1	0.80	138.4	0.76	113.3	0.81
11	181.5	0.80	151.2	0.82	168.3	0.80	138.0	0.82	112.3	0.85
12	179.0	1.10	160.0	0.81	172.0	0.83	138.1	0.84	--	--
13	182.0	0.82	167.0	0.83	179.0	0.83	142.3	0.88	116.8	0.82
14	191.0	0.83	174.0	0.83	191.0	0.84	151.0	0.86	121.0	0.83
15	204.0	0.85	183.0	0.88	203.6	0.85	158.6	0.80	135.5	0.90
16	221.6	0.87	191.2	0.86	218.3	0.87	177.3	0.89	152.5	0.89
17	234.0	0.88	203.0	1.00	235.3	0.88	--	0.86	171.0	0.92
18	248.0	0.90	213.0	0.87	258.3	0.84	210.0	0.92	190.0	0.93
19	257.0	0.90	226.1	0.84	266.5	0.90	223.4	0.94	210.5	0.96
20	264.0	0.90	--	0.87	279.4	1.00	240.8	0.94	236.5	0.94
21	278.0	0.92	250.0	0.90	301.0	0.92	246.0	--	243.0	0.96

[1] Respiratory rate in mg CO_2 per 100 g dry matter per day.

[2] Respiratory quotient: Ratio of carbon dioxide output to oxygen consumed.

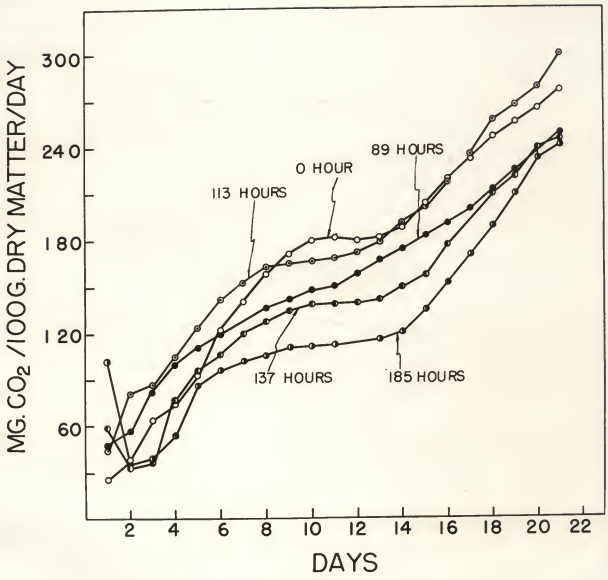


Fig. 6. Influence of dry-ice treatment interval and time on the respiration of wheat with 20.1 per cent moisture at 30-31° C.

Table 4. Exposure to dry ice treatment alone resulted in almost no change in the germinability of the wheat seeds up to 137 hours but the viability decreased regularly at longer treatment periods. Storage in respirometers for 21 days at 30-31° C. lowered the viability of all the samples even further, and this was obviously due to the extensive molding of the seeds under the conditions which were favorable to mold growth. There was a sharp increase in fat acidity of each sample as a result of the respiration trial. Different periods of dry ice treatment alone on the other hand had no significant effect on fat acidity as compared to that of the original wheat sample. As shown in Fig. 7, dry ice as the sole treatment of wheat with 20.1 per cent moisture content resulted in virtually no change in fluorescence or protein solubility; however, it did retard the normal increase in fluorescence which occurs when damp untreated wheat is stored in the respirometer for 21 days at 30-31° C. Protein solubility showed only a small decrease with storage under conditions favorable to fungal development. Dry ice treatment increased slightly the hydrating ability of the wheat gluten measured by the sedimentation test as the period of treatment increased but this value was lowered in all samples at the end of the respiration trial.

Effect of Gamma Radiation on Respiration, Germination, and Chemical Changes of Wheat Irradiated When Dry and Subsequently Wetted to 20 Per Cent Moisture Content

The data for the A series show that elimination of fungal activity in wheat at 19.8 per cent moisture content occurred at some dosage between 125,000 and 625,000 rep. Under these

Table 4. Moisture content, germination, fat acidity, fluorescence, per cent transmittancy (protein solubility), and sedimentation value of dry ice-treated wheat samples before and after the respiration trial of 21 days at 30-31° C.

Sample No.	Dry ice treatment: Time (hrs.)	Moisture content %: Initial	Moisture content %: Final	Germination %: Initial	Germination %: Final	Fat acidity MgKOH/100 g: Initial	Fat acidity MgKOH/100 g: Final	Fluorescence Scale units: Initial	Fluorescence Scale units: Final	Transmittancy %: Initial	Transmittancy %: Final	Sedimentation value[1]: Initial	Sedimentation value[1]: Final
B-1	Control (0)	20.1	20.8	96.0[2]	8.0	15.0	77.2	22.8	33.0	14.7	16.7	43.1	32.7
B-2	89	20.1	20.4	96.0	7.0	14.0	86.0	22.5	31.5	15.6	16.8	47.6	34.7
B-3	113	20.1	20.6	95.0	3.0	14.2	96.0	21.5	26.7	14.5	16.7	47.0	32.8
B-4	137	20.1	20.5	95.0	5.0	14.4	80.0	22.6	25.5	15.3	17.0	49.0	33.8
B-5	161	20.1	--	81.0	--	14.2	--	22.7	--	15.7	--	49.8	--
B-6	185	20.1	20.0	70.0	1.0	14.6	77.0	22.0	24.0	15.3	16.6	51.0	36.4

[1] 14 per cent moisture basis.

[2] Germination of original sample was 96 per cent.

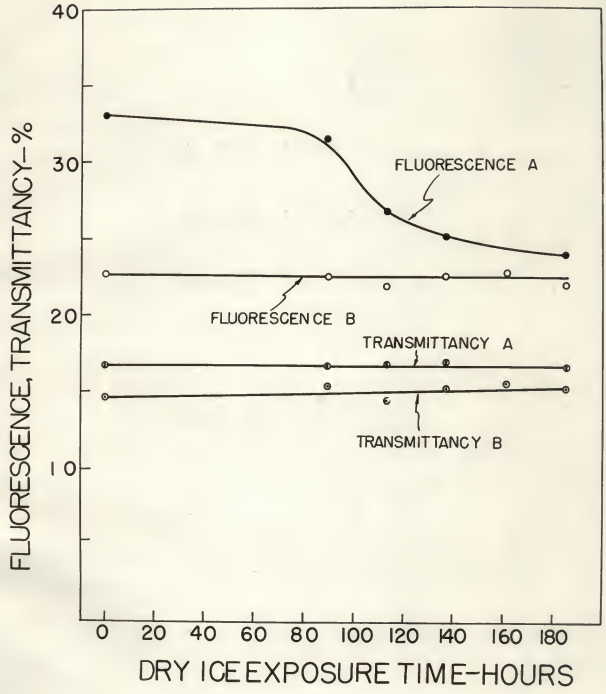


Fig. 7. Influence of dry-ice treatment interval on fluorescence and per cent transmittancy of extracts before (B) and after respiration trial (A) of 21 days at 30-31° C.

conditions seed viability was destroyed by the combined effect of radiation, dry ice treatment, and mold activity. In order to extend the study of the effect of moisture level on the effectiveness of gamma radiation, two sets of samples (each consisting of six samples) titled "C series" were conditioned to a moisture content of 12.4 per cent. These relatively dry samples, in contrast to A series, would not be expected to mold and thus did not require preservation in dry ice during shipment and handling. They were subjected to the same irradiation dosages as those of A series. Immediately on receipt in Manhattan, one set of the C series was wetted with distilled water under sterile conditions to 20 per cent moisture without removal of the sample from the container. This was accomplished by partial evacuation of the container with a water aspirator pump. This wetting was followed by thorough mixing and storage in the cold room overnight. The sample containers were then placed in the respiration apparatus (30-31° C.). The duplicate set of the C series was opened and chemical analysis on these samples was carried out immediately. Table 5 summarizes the respiratory data obtained with one set of the C series. As in A series, the respiratory quotients of samples C-5 and C-6 which received higher level of radiation dosages are greater than 1.0 excepting that of the first day of sample C-5, and in general, the respiratory quotients of sample C-6 are higher than those of sample C-4. The abnormally low values for respiratory quotients of sample C-3 were due to contamination with calcium chloride solution from the respirometer. Those of samples C-1, C-2, and C-3 appear to be characteristic of fungal activity. A graphical

Table 5. Influence of gamma radiation and time on the respiratory rate[1] and respiratory quotient[2] of wheat irradiated at a moisture content of 12.4 per cent.

	Sample No.											
	C-1		C-2		C-3		C-4[3]		C-5		C-6	
	Irradiation dosage rep											
	Control (0)		25,000		125,000		625,000		1,875,000		3,750,000	
Days	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.	CO_2	R.Q.
1	34.3	2.05	37.0	1.73	36.5	2.30	35.0	0.50	15.7	0.94	44.3	1.26
2	57.5	1.21	69.5	0.89	48.4	0.82	28.4	0.31	29.7	1.04	39.8	1.25
3	97.2	0.87	110.0	0.87	66.1	0.99	27.9	0.29	33.4	--	39.6	1.29
4	107.0	0.80	150.5	0.91	106.5	0.90	35.4	0.35	34.9	1.03	37.3	1.63
5	157.0	0.87	171.5	0.90	160.0	0.91	43.1	0.38	34.5	1.06	36.8	1.54
6	164.0	0.83	184.0	1.00	192.0	0.85	36.9	0.46	32.8	1.04	36.5	2.80
7	179.0	0.91	191.5	0.81	218.0	0.79	35.6	0.46	35.5	1.40	33.1	1.62
8	189.0	0.85	206.1	0.83	245.0	0.82	33.7	0.50	35.4	--	30.3	2.06
9	204.0	0.84	222.0	0.82	268.0	0.82	32.0	0.55	32.0	1.31	25.4	2.04
10	220.0	0.85	234.0	0.85	281.0	0.80	28.6	0.55	31.2	1.12	23.0	2.56

[1] Respiratory rate in mg CO_2 per 100 g dry matter per day.

[2] Respiratory quotient: Ratio of carbon dioxide output to oxygen consumed.

[3] Data for C-4 sample are anomalous, and there was evidence that it was contaminated with calcium chloride solutions from the spirometers.

presentation showing the influence of gamma radiation on daily carbon dioxide production is given in Fig. 8. Respiratory rate curves similar to those of the A series were obtained. With the irradiation of the wheat samples at a moisture content of 12.4 per cent, complete elimination of fungal activity was also attained at dosages of 625,000 rep and higher. However, the initial decrease in carbon dioxide production was absent in samples C-1, C-2, C-3, and C-5.

Table 6 is a summary of analytical results obtained from this series. Although these samples had been subjected to the same dosage as those of A series, the effect of radiation alone on wheat viability was less drastic. On the other hand, the change in sedimentation values was much greater, particularly at high dosage levels. Thus irradiation of wheat at 12.4 per cent moisture content seems to reduce the effect of gamma radiation in destroying seeds' viability. On the other hand, irradiation in a relatively dry state is more effective in decreasing the hydrating ability of wheat gluten progressively as the dosage level increased. It is apparent from Fig. 9 that similar trends in the development of fluorescence and changes in protein solubility occurred in comparison to those observed in the high-moisture A series.

Determination of More Precise Limits for Radiation Inhibition of Respiration in Dry Wheat and Accompanying Effects on Germination and Chemical Changes

Studies up to this point revealed that complete elimination of fungal activity in wheat at moisture content ranging from 12.4

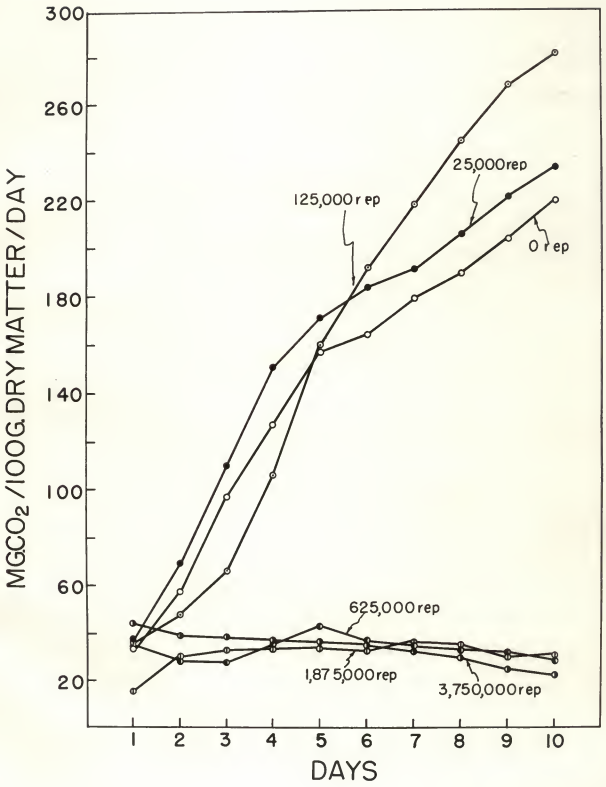


Fig. 8. Influence of gamma radiation and time on the respiration of wheat with 20 per cent moisture, irradiated at 12.4 per cent moisture.

Table 6. Influence of gamma radiation on germination, fat acidity, fluorescence, transmittancy (protein solubility), sedimentation value, and moisture content of C series before and after respiratory trial of 10 days at 30-31° C.

Sample No.	Irradiation dosage rep	Moisture content %		Germination %		Fat acidity MgKOH/100 g		Fluorescence Scale units		Transmittancy %		Sedimentation value[1]	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
C-1	Control (0)	20	20.3	96.0	9.0	16.0	72.0	20.0	25.0	18.0	15.8	48.0	46.4
C-2	25,000	20	20.3	92.0	7.0	15.0	72.0	20.0	24.4	16.6	15.5	45.1	42.4
C-3	125,000	20	20.0	70.0	4.0	16.0	63.4	20.2	24.0	16.3	17.8	39.1	43.0
C-4[2]	625,000	20	22.6	33.0	0.0	15.4	20.0	22.0	36.8	16.1	97.0	31.3	16.6
C-5	1,875,000	20	19.5	0.0	0.0	16.6	14.0	26.5	33.2	18.0	26.0	16.4	41.5
C-6	3,750,000	20	19.4	0.0	0.0	15.0	14.0	33.9	43.0	19.3	35.0	10.8	40.8

[1] 14 per cent moisture basis.

[2] Data for C-4 sample are anomalous, and there was evidence that it was contaminated with calcium chloride solution from respirometer.

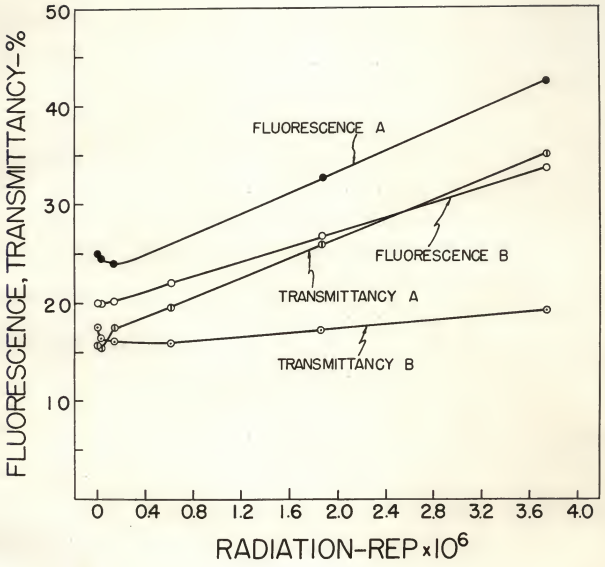


Fig. 9. Influence of gamma radiation on fluorescence and per cent transmittancy (protein solubility) of extracts of wheat before (B) and after respiration trial (A) of 10 days at 30-31° C.

per cent to 20 per cent occurred at some dosage between 125,000 and 625,000 rep. It seemed necessary to determine with greater precision the dosage required to destroy fungi in normal dry wheat. This investigation was carried out by setting up two sets of samples (D series) with three replicates in each set which were irradiated at a moisture level of 12.4 per cent with the following dosages:

Sample	Dosage rep
D-1	125,000
D-2	250,000
D-3	500,000

The usual sterile procedures were used to condition one set of the wheat seeds to 20 per cent moisture content upon receipt of the samples after radiation treatment. The other set of samples was analyzed immediately. The graphical data for the 12-day respiration trial appear in Fig. 10. It was noted that the daily carbon dioxide production of samples D-2 and D-3 showed a decreasing trend during the first several days, followed by a very slight regular increase during the remainder of the trial. In contrast, sample D-1 began with the lowest rate of carbon dioxide production which was maintained during the five days but showed a strong increase from the sixth day onward. Examination of the samples at the end of the trial revealed that only sample D-1 was moldy. The respiratory quotient values were in the range of 0.6 to 0.95 for all the samples.

Table 7 indicates that gamma radiation alone at the range from 125,000 to 500,000 rep has no significant effects on fat

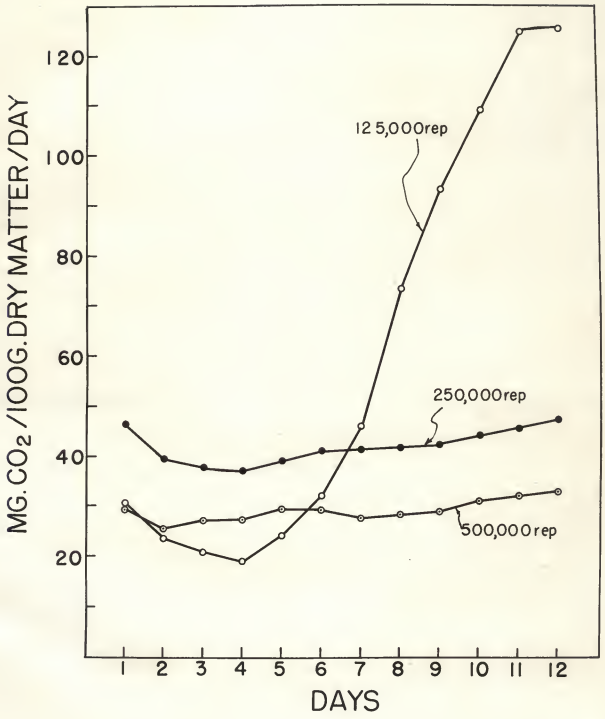


Fig. 10. Influence of gamma radiation in the critical range and time on the respiration of wheat with 20 per cent moisture, irradiated at 12.4 per cent moisture.

Table 7. Influence of gamma radiation on germination, fat acidity, fluorescence, per cent transmittancy (protein solubility), sedimentation value, and moisture content of D series before and after respiratory trial of 12 days at 30-31° C.

Sample No.	Irradiation dosage rep	Moisture content %		Germination[2] %		Fat acidity mgKOH/100 g		Fluorescence: Scale units		Transmittancy %		Sedimentation value[1]	
		Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
D-1	125,000	20	19.2	14.0	9.0	16.0	43.0	19.3	20.6	16.9	18.5	38.5	41.7
D-2	250,000	20	19.6	8.0	71.0	16.4	16.8	19.4	20.4	16.0	17.5	35.2	48.4
D-3	500,000	20	19.3	5.0	75.0	16.0	16.2	20.4	26.9	17.8	17.8	28.9	44.7

[1] The sedimentation value of D-0 (Control) is 44.5. Sedimentation value corrected to 14 per cent moisture.

[2] The germination test report from Kansas Seed Test Laboratory of these samples was completely anomalous.

acidity, fluorescence, and per cent transmittancy of all the samples. This fact has been pointed out in the studies with the A and C series. However, considerable increase in fat acidity of sample D-1 and the fluorescence of sample D-3 were observed at the termination of the 12-day respiration trial. Protein solubility, however, showed only a small increase for all the three samples. The high value of fat acidity of D-1 was clearly due to mold growth, and the high fluorescence of D-3 must be due to high radiation dosage. The germination test report from the Kansas Seed Test Laboratory of these three samples was completely anomalous and is not in agreement with those obtained previously in both A and C series. The trend of the change of sedimentation value with radiation dosage was noted to be the same as in C series. A graphical presentation of the development of fluorescence and change of protein solubility is given in Fig. 11. The above results indicate that with the material and conditions used in this experiment, the critical dose for elimination of fungi was 250,000 rep.

Evaluation of Effect of Gamma Radiation on Technological Properties of Wheat

The more precise evaluation of minimum treatment required to inhibit the fungal activity was found to be a dosage of 250,000 rep. Irradiation of wheat of 12.4 per cent moisture content with this dosage resulted in no apparent adverse effect in regard to development of fluorescence or alteration of protein solubility. However, information was still needed concerning the change in technological properties of wheat after given a dose sufficient to

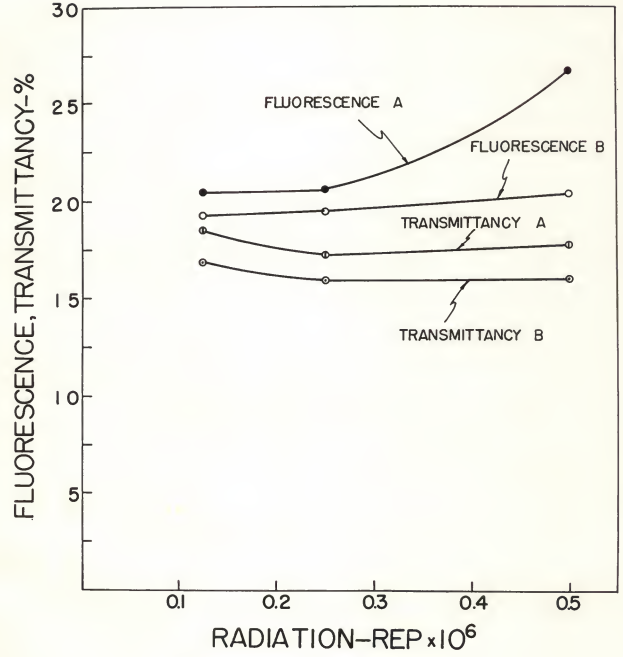


Fig. 11. Influence of gamma radiation on fluorescence and per cent transmittancy (protein solubility) of extracts of wheat before (B) and after respiration trial (A) of 12 days at 30-31° C.

eliminate the deteriorative fungi. This was accomplished by irradiating samples of the original wheat of 8.3 per cent moisture content to four different levels of disage, followed by a routine evaluation of milling and baking properties of the grain. Forty-eight No. 2 cans consisting of four separate lots, each of 12 cans, containing the wheat sample (each can contained approximately one pound of wheat) were shipped to the Quartermaster Food and Container Institute in Chicago which in turn shipped this material to Idaho Falls, Idaho for treatment in the Materials Testing Reactor there. This reactor delivers approximately 1.98×10^6 rep/hr. The following dosage treatments were applied at an ambient temperature of 72° F.

Sample	Radiation dosage rep	Radiation time hours	minutes	seconds
0	Control (0)	--	--	--
I	125,000	--	3	47
II	250,000	--	7	35
III	500,000	--	15	15
IV	1,000,000	--	30	18

Irradiation was completed on November 17. On the arrival of the treated samples in Manhattan on November 24, the cans were opened and the grain from all the 12 cans of each treatment was well mixed. The combined mixed samples were then conditioned to a moisture content of 15.5 per cent for milling by addition of a calculated amount of distilled water. Flours were prepared on the next day with the Buhler experimental mill. The following milling results were obtained.

Sample	Wheat wt. milled (g)	Bran		Shorts		Flour	
		Wt.(g)	%	Wt.(g)	%	Wt.(g)	%
0	5,180	1,180	22.8	810	15.6	2,960	57.0
I	6,220	1,460	23.5	840	13.5	3,690	59.4
II	6,340	1,440	22.7	750	11.8	3,900	61.5
III	6,250	1,445	23.1	795	12.7	3,610	57.8
IV	6,170	1,445	23.4	1,020	16.5	3,570	57.8

The data indicate no significant difference in milling properties among the various samples except for an unusually high yield of shorts from the most highly irradiated wheat.

The data for chemical analyses in Table 8 confirm the results obtained in both C and D series. There were no appreciable changes in fat acidity, fluorescence, and per cent transmittancy as the dosage increased from 0 to 1,000,000 rep. The sedimentation value, however, showed a significant decrease with radiation dosage.

Table 8. Influence of gamma radiation on germination, fat acidity, fluorescence, sedimentation value, and transmittancy of wheat irradiated at 8.3 per cent moisture content.

Sample	Irradiation dosage rep	Germination %	Fat acidity MgKOH/100g	Fluorescence scale units	Transmittancy %	Sedimentation value[1]
0	Control (0)	95.0	14.0	20.3	17.2	41.1
I	125,000	34.0	14.5	20.1	17.0	37.4
II	250,000	5.0	14.5	20.3	16.8	34.9
III	500,000	4.0	13.5	20.6	16.1	29.5
IV	1,000,000	1.0	14.0	20.9	16.6	21.8

[1] 14 per cent moisture basis.

One-pound regular commercial pan bread loaves were baked with these flours using a regular rich commercial formula, by a straight dough procedure. Each flour was tested for response to oxidation, using $KBrO_3$. The formula and method are described under Materials and Methods. Figure 12 shows dough mixing curves (Farinograms), prepared prior to baking in order to determine the absorption requirement as well as dough mixing characteristics. Plate I shows sections of loaves of bread made in the experiment. The data of Table 9 indicate that the absorption, i.e., water required to make a dough of optimum consistency, decreased slightly at 125,000 and 250,000 rep but with a dosage of 1,000,000 rep increased to a value more than 1 per cent greater than that of the untreated wheat. The dough development or mixing time shows a decrease with increasing dosage except for an anomalously high value in the case of 500,000 rep dosage. Mixing tolerance index in Brabender Units which is defined as the decreases in dough consistency from the maximum, and measured in arbitrary units at five minutes after initiation of mixing, showed a decrease at 125,000, 250,000, and 500,000 rep in comparison with untreated material. Dosage of 1,000,000 rep was accompanied by marked increases in M.T.I. indicating degradation of the physical structure of the dough. Loaf volume in the absence of potassium bromate ($KBrO_3$) showed an increase with all radiation dosages except 1,000,000 rep which was accompanied by a considerable loss in loaf volume. All loaves showed excellent response to potassium baromate; this reagent produces very fine bread even from flour treated with 1,000,000 rep.

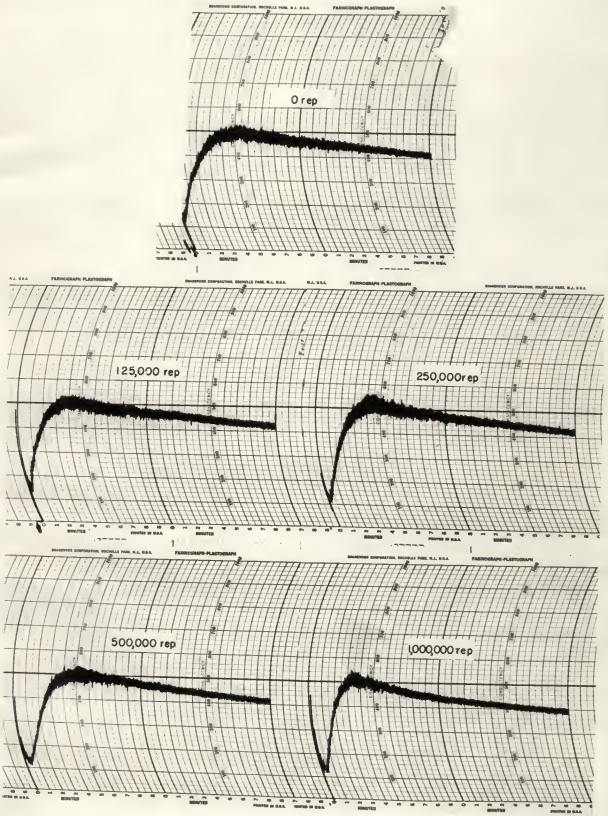


Fig. 12. Influence of gamma radiation on the farinograms (dough development curves) of flour prepared from irradiated wheat.

EXPLANATION OF PLATE I

Sections of bread loaves.

Sample No.	Irradiation dosage rep	$KBrO_3$ %
0-0	0	0.000
0-1	0	0.001
0-2	0	0.003
I-0	125,000	0.000
I-1	125,000	0.001
I-2	125,000	0.003
II-0	250,000	0.000
II-1	250,000	0.001
II-2	250,000	0.003
III-0	500,000	0.000
III-1	500,000	0.001
III-2	500,000	0.003
IV-0	1,000,000	0.000
IV-1	1,000,000	0.001
IV-2	1,000,000	0.003

PLATE I

Table 9. Farinograph and bread baking data.

Sample No.	Irradiation dosage rep	Flour moisture %	Flour protein %	Flour ash %	Farinograph absorption	Dough development time (min.)	M.T.I.[1] Brabender units	Loaf volume 0.000 %	Loaf volume 0.001 % KBrO3	Loaf volume 0.003 % KBrO3	Loaf score 0.000 %	Loaf score 0.001 % KBrO3	Loaf score 0.003 % KBrO3
0	Control (0)	12.90	13.8	0.39	71.14	5-1/2	40	2633	2962	3000+	82	92	94
I	125,000	13.20	13.6	0.37	70.08	4-1/4	30	2745	2970	3000+	83	93	95
II	250,000	13.53	13.8	0.37	70.94	4-1/2	24	2702	2972	3000+	79	89	88
III	500,000	13.65	13.9	0.37	71.90	5-1/8	30	2745	2907	3000+	78	84	85
IV	1,000,000	13.79	13.8	0.35	72.26	3-1/4	47	2490	2890	2938	66	73	73

[1] Mixing tolerance index.

The overall loaf score indicates that the best loaves were produced from wheat treated with 125,000 rep. Preliminary examination of the loaves showed no easy perceptible changes in flavor and odor.

DISCUSSION AND CONCLUSIONS

As shown in Figs. 3 and 8, elimination of fungal activity in wheat irradiated at widely different moisture values of 19.8 per cent and 12.4 per cent as indicated by subsequent levels of respiration, occurred at some dosage between 125,000 and 625,000 rep. It appears that the moisture content of wheat at irradiation (at least within this range) was not critical in regard to the efficiency of the gamma radiation in destroying the fungi or microorganism which are responsible for respiratory activity associated with the wheat grain.

The immediate initial decrease in carbon dioxide production shown in Fig. 3 and samples B-4 and B-6 in Fig. 6 and sample D-1 in Fig. 10 has been noted by other workers when dicotyledonous seed (18) and wheat germ (25) were freshly wetted. This apparently involves gas exchange due to certain respiratory enzymes of the seed rather than associated microorganisms. From comparison of Figs. 3, 6, 8, and 10, it can be seen that this initial level of carbon dioxide production and the succeeding decrease in respiration (Fig. 3) were related to the moisture content of the wheat at irradiation, the time intervals required at room temperature to expose the wheat samples of high dosage level to irradiation, the radiation dosage applied, and the exposure to low temperature.

Thus from Fig. 6 it appears that holding damp grain at low temperature favors initial high levels in the preliminary decrease of carbon dioxide production at 30-31° C. This would suggest inhibition of an enzyme causing decarboxylation in the seed embryos. When molds are the principal contributors to the respiration, the respiratory quotients of damp wheat are in the range of 0.8 to 1.0 (values higher than 1.0 occurred in the first day of the dry ice treated sample and those samples irradiated at 12.4 per cent moisture and subsequently wetted to 20 per cent moisture). High respiratory quotient values occurred in heavily-irradiated samples A-5, A-6 in Fig. 5 and samples C-5, C-6 in Table 5. This suggests that high dosage of ionizing radiation may decrease the oxygen uptake ability of wheat seeds without hindering the breakdown of polycarboxylic respiratory intermediate (25).

Irradiation of wheat seeds alone under both damp and relatively dry conditions resulted in no significant change in fat acidity of the seeds. This fact has also been reported for cottonseed by Lambou, et al. (21) in a study of effects of high-voltage cathode-ray irradiation. Different periods of dry ice treatment were also without effect on fat acidity. It seemed that ionizing radiation or dry ice treatment does not produce hydrolysis of fat. Considerable increase in fat acidity occurred in samples which were given insufficient amounts of radiation to eliminate the fungal respiration and this obviously was due to lipase produced by mold (Tables 2, 4, 6, and 7).

Exposure of wheat seeds to dry ice treatment alone for a period up to 137 hours, resulted in no change in germinability, but

seed viability decreased regularly at treatment periods longer than 137 hours (Table 4). Irradiation of wheat at 19.8 per cent moisture together with eight days of exposure to dry ice treatment during shipment and handling, resulted in greater change in seed viability than irradiation of wheat in a relatively dry state of 12.4 per cent moisture (Tables 2 and 6). It seems that the viability of wheat seeds of 19.8 per cent moisture subjected to dry ice treatment was more sensitive to gamma radiation than that of wheat of 12.4 per cent moisture. There was no simple relationship between the extent of survival of viable seed following radiation treatment and subsequent respiratory activity due to seed and/or fungi. Sample C-3, for example, showed nearly a one-third reduction in seed viability due to irradiation as compared to that of the control sample, yet the subsequent fungal respiration is not inhibited. In sample C-4, with two-thirds loss of seed viability, the fungal activity was apparently destroyed (Table 6). Complete loss of viability occurred when wheat was given a dose of 1,875,000 rep at 12.4 per cent moisture content, and this critical value is very close to 2,000,000 rep of cathode-ray dosage required to destroy the viability of cottonseed as reported by Lambou, et al. (21). The viability of seed samples in which radiation was not enough to inhibit fungal deterioration, was drastically lowered as a result of respiration trial. This was obviously due to the toxic action of products of mold growth.

A change of fluorescence probably indicates some alteration in the protein portion of wheat seeds, and may be considered a very sensitive indicator of change in biological material.

Radiation of wheat at 19.8 per cent moisture together with eight days of dry ice treatment resulted in no appreciable change of fluorescence in samples which had been given doses insufficient to eliminate the deterioration due to fingi (i.e., from 0 to 125,000 rep). However, an increase in fluorescence did occur in samples which received a dosage of 625,000 rep or more. In samples in which radiation dosage was insufficient to eliminate fungi, storage in dry ice followed by respiration trial for 17 days at 30-31° C. caused the fluorescence to show a slight increase. However, a remarkable increase occurred also in samples in which no fungal respiration was evident. Moisture content of wheat at the time of irradiation had no significant effect on development of fluorescence in samples in which radiation was not enough to inhibit the fungal activity. However, ionizing radiation of wheat in a relatively dry state (8.3 and 12.4 per cent moisture) favors the development of fluorescence in heavily irradiated samples. Different periods of dry ice treatment alone had no effect on the development of fluorescence. However, the dry ice treatment retarded the normal increase in fluorescence which occurs when damp untreated wheat is stored in the respirometer.

Protein solubility, as indicated by the transmittancy value, is an estimation of colloidal water-binding property of the protein. Protein solubility increased slightly with radiation dosages up to 625,000 rep under both damp (19.8 per cent moisture 8 days dry ice treatment) and dry conditions (moisture content ranging from 12.4 to 8.3 per cent). A slight increase occurred at higher dosages. It seems that at low dosage levels, ionizing radiation

increases the water-binding property of protein but that this effect is lost at more elevated dosage levels. Considerable decrease in protein solubility occurred in heavily irradiated samples in which no mold growth occurred after respiration trial. Because these samples were not moldy, this continued decrease in protein solubility must be due to chemical changes in the seeds when maintained at elevated temperature and moisture content. The increased fluorescence exhibited by these samples may be an indication that initial phases of the browning reaction (i.e., interaction of protein and carbohydrates) are taking place.

Sedimentation value, also a measure of the hydrating potential of protein, decreased significantly and regularly with dosage from 0 to 3,750,000 rep when wheat was irradiated at 12.4 per cent moisture content (Table 6). At the end of the respiration trial, a slight decrease in sedimentation value occurred in samples C-1 and C-2 given doses of insufficient amount to prevent the growth of molds, but samples in which no fungal activity occurred, showed a remarkable increase, particularly at higher dosage levels. The phenomenon was quite in contrast to the development and change in protein solubility, and is difficult to explain for the time being. The combined effect of irradiation of wheat at 19.8 per cent moisture together with eight days of dry ice treatment, caused no appreciable variation in sedimentation value in samples given doses from 0 to 1,875,000 rep. However, an increase in sedimentation value occurred in the sample which received the highest dosage of 3,750,000 rep. Nevertheless, the increase in sedimentation value after holding these high-moisture, dry ice treated, heavily

irradiated samples in the respirometer was much less pronounced than with those irradiated at a relatively dry condition (Tables 2 and 6). It seems that the relatively damp condition of wheat and eight days dry ice treatment prevents a change in the protein hydrating ability caused by irradiation. This point is apparent from the results of dry ice treatment alone on sedimentation value. Table 4 indicates that the longer the time of exposure to dry ice, the greater the sedimentation value. However, in contrast to the irradiated samples, the sedimentation value for each of these samples receiving only dry ice treatment was lowered as a result of the respiration trial (Table 4).

Figure 10 shows that elimination of fungal activity occurred when wheat was given a dose of 250,000 rep at 12.4 per cent moisture. No adverse effect in regard to development of fluorescence and change in protein solubility accompanied this radiation treatment. About six points of decrease in sedimentation value was observed (Table 8). No significant change in milling properties occurred in wheat irradiated at 8.3 per cent moisture and with a dosage range from 0 up to 1,000,000 rep, except the most heavily irradiated sample showed an unusually high yield of shorts. The absorption, i.e., water required to make a dough of optimum consistency, decreased slightly at 125,000 and 250,000 rep but increased at high radiation levels. This dryness of irradiated flour has been noted by Brownell, et al. (8). Dough development time decreased regularly except for an anomalously high value in the case of 500,000 rep dosage. This may be an indication of a breakdown of protein molecules by irradiation. The high M.T.I.

value in a heavily irradiated sample suggests that the ionizing radiations damaged the protein and thus in turn the dough structure. Loaf volume in the absence of potassium bromate ($KBrO_3$) increases with dosage up to 500,000 rep, and thus the decrease of sedimentation value had no effect on loaf volume. It was also noted that all loaves showed excellent response to potassium bromate even when the flour had very low sedimentation value due to irradiation treatment. The baking quality of wheat as determined by total loaf score was increased with a radiation of 125,000 rep but decreased regularly at high doses. Loaves made with flours prepared from wheat given 250,000, 500,000, and 1,000,000 rep had darker crumb color and coarser grain and texture. Brownell, et al. (8) reported that bread made with flours after direct irradiation with doses of 200,000 and 500,000 rep resulted in smaller volume in comparison with the loaf made with non-irradiated flour. In the present case, this indicates that the increase in loaf volume observed with treatment beyond 125,000 rep occurred at the expense of other factors such as crumb color, grain, and texture. The data suggest that radiation treatment of dry wheat can improve the baking properties even though some breakdown or modification of protein structure has occurred as indicated by a lowering of the sedimentation value. It is well known that some modification of gluten protein by proteolytic enzymes during the dough fermentation process in baking is desirable in terms of improved bread characteristics. It appears from these studies that such a desirable modification can be achieved by ionizing radiations which at the same time would improve the storage properties of the grain

by eliminating insect life and some of the microorganisms responsible for deterioration. Treatment of dry wheat with radiations to a level which produces these desirable effects apparently causes no other undesirable biochemical change (as indicated by fluorescence, protein solubility, and fat acidity) except for a drastic loss in seed viability.

SUMMARY

A study was carried out on the effect of gamma radiation treatment of wheat on the subsequent respiratory activity, which at elevated moisture contents, is primarily due to fungi. Irradiations were applied under both damp and relatively dry conditions. Daily measurement of oxygen consumption and carbon dioxide production were carried out over prolonged time intervals. Chemical and biological changes of wheat accompanying the radiation treatment alone were determined as well as the combined effect of radiation, dry ice treatment, and prolonged storage in respirometers. The investigation was completed with an evaluation of the influence of gamma radiation on the technological (milling and baking) properties of wheat. The following results were obtained.

Moisture content of wheat ranging from 19.8 to 12.4 per cent at irradiation had no effect on the efficiency of gamma radiation in destroying fungi or other microorganisms associated with the storage deterioration of wheat grain, as indicated by the subsequent levels of respiration.

Complete elimination of fungal respiration in this range of

moisture content was found to be at some dosage between 125,000 and 625,000 rep. A characteristic initial release of carbon dioxide over a period of a few days and a succeeding decrease in gas exchange, prior to appearance of fungal were related to the moisture content of the wheat at irradiation, the time interval required at room temperature to expose the wheat samples of high dosage level to irradiation, the radiation dosage applied, and the exposure to low temperature.

The longer the exposure to dry ice, the lower was the maximum production of carbon dioxide due to fungi.

When molds are the principal contributors to the respiration, the respiratory quotients of damp wheat are in the range of 0.8 to 1.0. Much higher respiratory quotient values occurred in heavily-irradiated wheat samples during the initial period of carbon dioxide release due to non-fungal agencies, as well as in the first day after treatment of damp wheat to low temperature by means of dry ice.

Following either radiation or dry ice treatment when applied singly, no significant change in fat acidity occurred when wheat was irradiated under both damp and relatively dry conditions (moisture content ranging from 19.8 to 12.4 per cent) from doses of 25,000 to 3,750,000 rep or dry ice treatment for a period of 0 to 185 hours. However, when respiration trials followed such irradiation or dry ice treatment, considerable increase in fat acidity occurred in samples in which radiation was not sufficient to inhibit the fungi, and in those which had been exposed to dry ice treatment alone.

Exposure of wheat seeds to dry ice treatment alone for intervals up to 137 hours resulted in no change in seed viability, but the germination decreased regularly at treatment periods longer than 137 hours.

Gamma radiation was more effective in destroying seed viability when the wheat contained 19.8 per cent moisture content and was exposed to an eight day dry ice treatment, than when the grain was irradiated at 12.4 per cent moisture content. Irradiation of wheat at 12.4 per cent moisture content with a dose of 1,875,000 rep caused complete loss of seed viability.

Irradiation of wheat under both damp and relatively dry conditions, caused no appreciable variation in fluorescence of samples which had been given doses up to 125,000 rep. However, fluorescence increased regularly at doses higher than 125,000 rep. Relatively little increase of fluorescence occurred after respiratory trial in samples in which there was mold growth. On the other hand, samples which received sufficient radiation to eliminate fungi showed no respiratory increase but did exhibit considerable increases in fluorescence. The ionizing radiations were more effective in producing fluorescence changes when the wheat was irradiated under relatively dry conditions particularly at high dosage values. Dry ice treatment alone (without irradiation) resulted in no significant change in fluorescence, however, this treatment retarded the normal increase in fluorescence which occurs when damp untreated wheat is stored in the respirometer.

Protein solubility following irradiation of wheat at 19.8 and 12.4 per cent moisture content increased slightly with

radiation dosages up to 625,000 rep and decreased a little at higher dosage values. At the end of the respiratory trials, there was no appreciable decrease in protein solubility in samples in which irradiation was not sufficient to inhibit the fungal respiration. However, a remarkable increase in protein solubility occurred in samples where no mold activity existed. Moisture content of wheat (from 19.8 to 8.3 per cent at irradiation) had no significant effect on protein solubility. Exposure of wheat seeds to dry ice treatment alone had no effect on protein solubility, but a slight decrease in protein solubility occurred as a result of fungal growth on such grain.

Irradiation of wheat alone at 12.4 per cent moisture content decreased the sedimentation values markedly as dosage increased from 0 to 3,750,000 rep, although at the end of the respiration trial, a slight decrease in sedimentation value occurred in samples which received 0 and 25,000 rep, respectively. On the other hand, non-moldy samples which received doses higher than 125,000 rep showed a considerable increase in sedimentation value. It was also noted that moisture content of wheat at irradiation had no appreciable effect on sedimentation value. Following the radiation of wheat at 19.8 per cent moisture together with eight days of dry ice treatment, there was no significant variation in sedimentation values in samples given doses from 0 to 1,875,000 rep but there was an increase in the sample which received the highest dosage of 3,750,000 rep. Little increase in sedimentation value occurred in the radiation and dry ice treated samples except in the most heavily irradiated one in which no clear sediment boundary could

be observed. Dry ice treatment of wheat alone caused a slight increase in sedimentation value as the treatment period increased from 0 up to 185 hours. A slight decrease in this value occurred in each sample after the respiration trial.

The minimum dosage required to inhibit the fungal activity in dry wheat (12.4 per cent moisture) was found to be 250,000 rep. Although no adverse effect in regard to development of fluorescence or change in protein solubility was observed due to the radiation, a slight decrease in sedimentation value did occur. Nevertheless, the loaf volume of the bread made with flour prepared from wheat irradiated with this dosage was greater than that of untreated sample. However, the loaf total score was decreased slightly in comparison with the untreated sample due to a somewhat coarser crumb grain and texture.

The baking quality of dry wheat was improved by irradiation with a dose of 125,000 rep of gamma radiation but was lowered by higher doses. Potassium bromate caused radiation-damaged flour to yield loaf volumes comparable to non-irradiated material. No significant change in milling properties was observed in wheat given doses up to 1,000,000 rep.

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INFLUENCE OF RADIATION STERILIZATION ON RESPIRATION AND OTHER PROPERTIES OF DORMANT WHEAT SEEDS

by

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A study was carried out on the effect of gamma radiation treatment of wheat on the subsequent respiratory activity, which at elevated moisture contents, is primarily due to fungi. Irradiations were applied under both damp and relatively dry conditions. Daily measurement of oxygen consumption and carbon dioxide production were carried out over prolonged time intervals. Chemical and biological changes of wheat accompanying the radiation treatment alone were determined as well as the combined effect of radiation, dry ice treatment, and prolonged storage in respirometers. The investigation was completed with an evaluation of the influence of gamma radiation on the technological (milling and baking) properties of wheat. The following results were obtained.

Moisture content of wheat ranging from 19.8 to 12.4 per cent at irradiation had no effect on the efficiency of gamma radiation in destroying fungi or other microorganisms associated with the storage deterioration of wheat grain, as indicated by the subsequent levels of respiration.

Complete elimination of fungal respiration in this range of moisture content was found to be at some dosage between 125,000 and 625,000 rep. A characteristic initial release of carbon dioxide over a period of a few days and a succeeding decrease in gas exchange, prior to appearance of fungi were related to the moisture content of the wheat at irradiation, the time interval required at room temperature to expose the wheat samples of high dosage level to irradiation, the radiation dosage applied, and the exposure to low temperature.

The longer the exposure to dry ice, the lower was the maximum production of carbon dioxide due to fungi.

When molds are the principal contributors to the respiration, the respiratory quotients of damp wheat are in the range of 0.8 to 1.0. Much higher respiratory quotient values occurred in heavily-irradiated wheat samples during the initial period of carbon dioxide release due to non-fungal agencies, as well as in the first day after treatment of damp wheat to low temperature by means of dry ice.

Following either radiation or dry ice treatment when applied singly, no significant change in fat acidity occurred when wheat was irradiated under both damp and relatively dry conditions (moisture content ranging from 19.8 to 12.4 per cent) from doses of 25,000 to 3,750,000 rep or dry ice treatment for a period of 0 to 185 hours. However, when respiration trials followed such irradiation or dry ice treatment, considerable increase in fat acidity occurred in samples in which radiation was not sufficient to inhibit the fungi, and in those which had been exposed to dry ice treatment alone.

Exposure of wheat seeds to dry ice treatment alone for intervals up to 137 hours resulted in no change in seed viability, but the germination decreased regularly at treatment periods longer than 137 hours.

Gamma radiation was more effective in destroying seed viability when the wheat contained 19.8 per cent moisture content and was exposed to an eight day dry ice treatment, than when the grain was irradiated at 12.4 per cent moisture content. Irradiation of

wheat at 12.4 per cent moisture content with a dose of 1,875,000 rep caused complete loss of seed viability.

Irradiation of wheat under both damp and relatively dry conditions, caused no appreciable variation in fluorescence of samples which had been given doses up to 125,000 rep. However, fluorescence increased regularly at doses higher than 125,000 rep. Relatively little increase of fluorescence occurred after respiratory trial in samples in which there was mold growth. On the other hand, samples which received sufficient radiation to eliminate fungi showed no respiratory increase but did exhibit considerable increases in fluorescence. The ionizing radiations were more effective in producing fluorescence changes when the wheat was irradiated under relatively dry conditions, particularly at high dosage values. Dry ice treatment alone (without irradiation) resulted in no significant change in fluorescence, however, this treatment retarded the normal increase in fluorescence which occurs when damp untreated wheat is stored in the respirometer.

Protein solubility following irradiation of wheat at 19.8 and 12.4 per cent moisture content increased slightly with radiation dosages up to 625,000 rep and decreased a little at higher dosage values. At the end of the respiratory trials, there was no appreciable decrease in protein solubility in samples in which irradiation was not sufficient to inhibit the fungal respiration. However, a remarkable increase in protein solubility occurred in samples where no mold activity existed. Moisture content of wheat (from 19.8 to 8.3 per cent at irradiation) had no significant

effect on protein solubility. Exposure of wheat seeds to dry ice treatment alone had no effect on protein solubility, but a slight decrease in protein solubility occurred as a result of fungal growth on such grain.

Irradiation of wheat alone at 12.4 per cent moisture content decreased the sedimentation values markedly as dosage increased from 0 to 3,750,000 rep, although at the end of the respiration trial, a slight decrease in sedimentation value occurred in samples which received 0 and 25,000 rep, respectively. On the other hand, non-moldy samples which received doses higher than 125,000 rep showed a considerable increase in sedimentation value. It was also noted that moisture content of wheat at irradiation had no appreciable effect on sedimentation value. Following the radiation of wheat at 19.8 per cent moisture together with eight days of dry ice treatment, there was no significant variation in sedimentation values in samples given doses from 0 to 1,875,000 rep but there was an increase in the sample which received the highest dosage of 3,750,000 rep. Little increase in sedimentation value occurred in the radiation and dry ice treated samples except in the most heavily irradiated one in which no clear sediment boundary could be observed. Dry ice treatment of wheat alone caused a slight increase in sedimentation value as the treatment period increased from 0 up to 185 hours. A slight decrease in this value occurred in each sample after the respiration trial.

The minimum dosage required to inhibit the fungal activity in dry wheat (12.4 per cent moisture) was found to be 250,000 rep. Although no adverse effect in regard to development of fluorescence

or change in protein solubility was observed due to the radiation, a slight decrease in sedimentation value did occur. Nevertheless, the loaf volume of the bread made with flour prepared from wheat irradiated with this dosage was greater than that of the untreated sample. However, the loaf total score was decreased slightly in comparison with the untreated sample due to a somewhat coarser crumb grain and texture.

The baking quality of dry wheat was improved by irradiation with a dose of 125,000 rep of gamma radiation but was lowered by higher doses. Potassium bromate caused radiation-damaged flour to yield loaf volumes comparable to non-irradiated material. No significant change in milling properties was observed in wheat given doses up to 1,000,000 rep.